CLAIMS

1. Method of diagnosing in an individual recent exposure to an agent which is a pathogen, vaccine or any other moiety which induces a cellular response, said method comprising determining in vitro whether the T cells of the individual recognise a protein from said agent having a length of at least 30 amino acids, to a greater extent than one or more peptide epitopes from the agent, a greater extent of recognition of the protein indicating that the individual has recently been exposed to the agent.

2. Method according to claim 1 comprising determining whether T cells of the individual exhibit a greater reaction to a protein from said agent having a length of at least 30 amino acids than to one or more peptide epitopes from the agent, a greater reaction indicating that the individual has recently been exposed to the agent.

3. Method according to claim 1 wherein determining whether the T cells recognise said protein is performed by determining the reaction of the T cells to an analogue of the protein which is recognised by T cells which recognise said protein, wherein said analogue has a length of at least 30 amino acids.

4. Method according to claim 1 or 3 wherein determining whether the T cells recognise said peptide epitope is performed by determining the reaction of the T cells to an analogue of the peptide epitope which analogue is recognised by T cells which recognise said peptide epitope.

5. Method according to any one of the preceding claims comprising:
(i) contacting a first population of T cells from the individual with (a) one or more peptide epitopes from the agent, or (b) an analogue of said peptide(s) which is recognised by T cells that recognise said peptide(s), and determining the reaction of the T cells to the peptide(s) or analogue(s), and
(ii) contacting a second population of T cells from the individual with (a) a protein from the agent, or (b) an analogue of said protein which is recognised by T cells that recognise said protein, wherein the protein or analogue has a length of at least 30 amino acids and determining the reaction of the T cells to the protein or analogue.

6. Method according to any one of the preceding claims in which the individual is diagnosed as having been exposed to the agent recently if there is substantially no reaction of the T cells to the peptide epitope or an analogue thereof.

7. Method according to any one of the preceding claims in which protein or its analogue comprises at least the amino acid sequence of the peptide epitope or its analogue.

8. Method according to any one of the preceding claims in which the peptide epitope, or the analogue of the peptide epitope, has a length of 8 to 29 amino acids.

9. Method according to any one of the preceding claims wherein whether or not the T cells recognise a pool of a least 4 peptide epitopes, or analogues thereof, is determined.

10. Method according to any one of the preceding claims wherein a pool of peptide epitopes and/or analogues which together represent all of the possible epitopes from the protein is used.

11. Method according to any one of the preceding claims in which during detection of the reaction of the T cells to the protein, or the analogue of the protein, antigen presenting cells are present which are capable of processing the protein and presenting it to the T cells.

12. Method according to any one of the preceding claims wherein the pathogen is an intracellular pathogen or the vaccine is against an intracellular pathogen.

13. Method according to any one of the preceding claims wherein the pathogen is HPV, HIV, SIV, HCV, a Chlamydia species, HBV, EBV, CMV, VZV, HSV, Legionella, S. typhi, P. falciparum, Leishmaniasis, M. leprae, influenza virus, foot and mouth virus, a Toxoplasma species, a Brucella species, a Cryptococcus species, a Candida species or an Aspergillus species; or the vaccine is against any of these

pathogens.

14. Method according to any one of claims 1 to 12 wherein the pathogen is M. tuberculosis or the vaccine is against M. tuberculosis.

15. Method according to any one of the preceding claims wherein the protein and/or epitope peptide is from ESAT-6 or CFP10.

16. Method according to any one of the preceding claims wherein the peptide(s) is chosen from one or more of the following peptide epitopes:

MTEQQWNFAGIEAAA
WNFAGIEAAASAIQG
IEAAASAIQGNVTSI
SAIQGNVTSIHSLLD
NVTSIHSLLDEGKQS
HSLLDEGKQSLTKLA
EGKQSLTKLAAAWGG
LTKLAAAWGGSGSEA
AAWGGSGSEAYQGVQ
SGSEAYQGVQQKWDA
YQGVQQKWDATATEL
QKWDATATELNNALQ
TATELNNALQNLART
NNALQNLARTISEAG
NLARTISEAGQAMAS
ISEAGQAMASTEGNV
QAMASTEGNVTGMFA
MAEMKTDAATLAQEA
TDAATLAQEAGNFER
LAQEAGNFERISGDL
GNFERISGDLKTQID
ISGDLKTQIDQVEST

KTQIDQVESTAGSLQ
QVESTAGSLQGQWRG
AGSLQGQWRGAAGTA
GQWRGAAGTAAQAAV
AAGTAAQAAVVRFQE
AQAAVVRFQEAANKQ
VRFQEAANKQKQELD
AANKQKQELDEISTN
KQELDEISTNIRQAG
EISTNIRQAGVQYSR
IRQAGVQYSRADEEQ
VQYSRADEEQQQALS
ADEEQQQALSSQMGF

or an analogue thereof which is recognised by a T cell which recognises the peptide epitope.

17. Method according to any one of the preceding claims wherein recognition of the peptide epitope, or its analogue, or of the protein, or its analogue, is determined by detecting secretion of a cytokine from the T cells.

18. Method according to claim 17 in which the cytokine is IFN-(.

19. Method according to claim 17 or 18 in which the cytokine is detected by allowing the cytokine to bind to an immobilised antibody specific to the cytokine and then detecting the presence of the antibody/cytokine complex.

20. Method of diagnosing in an individual recent exposure to an agent which is pathogen, vaccine or any other moiety which induces a cellular response, said method comprising determining in vivo whether the T cells of the individual recognise a protein from said agent having a length of at least 30 amino acids, to a greater extent than a peptide epitope from the agent, a greater extent of recognition of the protein indicating that the individual has recently been exposed to the agent.

21. Method according to claim 20 which is performed using the method of any one of claims 2 to 19.

22. Use a protein having a length of at least 30 amino acids from an agent which is a pathogen, vaccine or any other moiety which induces a cellular response and/or one or more peptide epitopes from the agent in the manufacture of a diagnostic means for use in a method of diagnosing in an individual recent exposure to the agent, said method comprising determining whether the T cells of the individual recognise the protein to a greater extent than peptide epitope(s), a greater extent of recognition of the protein indicating that the individual has recently been exposed to the agent.

23. A product comprising a protein from an agent which is a pathogen, vaccine or any other moiety which induces a cellular response, said protein having a length of at least 30 amino acids, and/or one or more peptide epitopes from the agent for separate, simultaneous or sequential use in a method of diagnosing in an individual recent exposure to the agent, said method comprising determining whether the T cells of the individual recognise the protein to a greater extent than the peptide epitope(s), a greater extent of recognition of the protein indicating that the individual has recently been exposed to the agent.

24. Method of treating an individual comprising administering to an individual diagnosed as having been exposed recently to a pathogen by a method according to any of the preceding claims, a product which prevent or treats the condition caused by the pathogen.

25. Use of a product which prevents or treats a condition caused by a pathogen in the manufacture of a medicament for the therapy of an individual who has been diagnosed as having been recently exposed to the pathogen by a method according to any one of claims 1 to 21.

26. Method or use according to claim 24 or 25 wherein the pathogen is M. tuberculosis

and/or the agent is rifampicin, isoniazid, pyrazinamide, ethambutol, streptomycin, para-amino-salicyclic acid, kanamyin, capreomycin, ethionamide, cycloserine, thiacetazone or a flouroquinolone, or an analogue of such an agent.

27. A kit for carrying out the method of any one of claims 1 to 21 comprising (i) said epitope peptide or said analogue thereof, and (ii) said protein or said analogue thereof, and optionally also a means to detect whether T cells recognise (i) and (ii).

28. A kit according to claim 27 which also comprises an agent as defined in any one of claims 24 to 26.

29. Method of diagnosing an individual who has cleared an infection by a pathogen comprising determining whether the T cells of the individual recognise antigen from the pathogen at a first and a subsequent second time point after exposure to the pathogen, wherein the finding that the T cells recognise antigen at the first time point and not at the second time point indicates that the individual has cleared the infection.

30. Method of diagnosing an individual who is more likely to progress to active disease after exposure to a pathogen comprising determining whether the T cells of the individual recognise antigen from the pathogen at a first and subsequent second time point after exposure to the pathogen, wherein the finding that the T cells do not recognise the antigen at the first time point, but do recognise the antigen at the second time point indicates that the individual is more likely to progress to active disease.

31. Method of diagnosing an individual who mounts a weaker response to a vaccine or a moiety which induces a cellular response after exposure to the vaccine or moiety comprising determining whether the T cells of the individual recognise antigen from the vaccine or moiety at a first and subsequent second time point after exposure, wherein the finding that the T cells do not recognise the antigen at the first time point, but do recognise the antigen at the second time point indicates that the individual is mounting a weaker response to the vaccine or moiety.

32. Method of testing the efficacy of a vaccine which has been administered to an individual comprising determining whether the T cells of the individual recognise antigen from the pathogen at a first and a subsequent second time point after exposure to the pathogen, wherein the finding that the T cells recognise antigen at the first time point and not at the second time point indicates that the vaccine antigen has been cleared and is not persisting.

33. Method according to any one of claims 29 to 32 wherein the first time point and second time point are separated by about 12 weeks.

34. Method according to any one of claims 29 to 33 wherein the first time point is about 2 to 16 weeks after exposure and/or the second time point is about 18 to 48 weeks after exposure.

35. Method according to any one of claims 29 to 34 wherein said determining is performed by detecting whether the T cells of the individual recognise/react to an antigen from said pathogen, vaccine or moiety.

36. Method according to any one of claims 29 to 35 wherein the antigen is a protein from said pathogen, vaccine or moiety having a length of at least 30 amino acids or is a peptide epitope from the pathogen, vaccine or moiety, wherein the protein or peptide epitope are as defined in any one of the preceding claims.

37. Method according to any one of claims 29 to 36 wherein said determining is performed by detecting the reaction of the T cells to an analogue of antigen which is optionally an analogue as defined in any one of the preceding claims.

38. Method according to any one of claims 29 to 37 wherein said determining is performed by contacting in vitro or in vivo a population of T cells from the individual with the antigen, or an analogue of said antigen and determining the reaction of the T cells to the antigen or analogue.

39. Method according to any one of claims 29 to 38 wherein the determination is deemed negative if there is substantially no reaction of the T cells to the antigen or analogue.

40. Method according to any one of the claims 29 to 39 wherein said determining comprises detecting whether or not the T cells recognise a pool of a least 4 peptide epitopes, or analogues thereof.

41. Method according to any one of claims 29 to 40 wherein during said determining antigen presenting cells are present which are capable of processing the antigen or analogue and presenting it to the T cells.

42. Method according to any one of claims 29 to 41 wherein the pathogen is an intracellular pathogen or the vaccine is against an intracellular pathogen.

43. Method according to any one of claims 29 to 42 wherein the pathogen is HPV, HIV, SIV, HCV, a Chlamydia species, HBV, EBV, CMV, VZV, HSV, Legionella, S. typhi, P. falciparum, Leishmaniasis, M. leprae, influenza virus, foot and mouth virus, a Toxoplasma species, a Brucella species, a Cryptococcus species, a Candida species or an Aspergillus species; or the vaccine is against any of these pathogens.

44. Method according to any one of claims 29 to 42 wherein the pathogen is M. tuberculosis or the vaccine is against M. tuberculosis.

45. Method according to any one of claims 29 to 44 wherein recognition of the antigen or analogue is determined by detecting secretion of a cytokine from the T cells.

46. Method according to claim 45 in which the cytokine is IFN-(.

47. Method according to claim 45 or 46 in which the cytokine is detected by allowing the cytokine to bind to an immobilised antibody specific to the cytokine and then

detecting the presence of the antibody/cytokine complex.

48. Method of treating an individual comprising administering to an individual diagnosed as being infected by a pathogen by a method according to any of claims 30 to 47, a product which prevent or treats the condition caused by the pathogen.

49. Method according to claim 48 wherein the pathogen is M. tuberculosis and/or the agent is rifampicin, isoniazid, pyrazinamide, ethambutol, streptomycin, para-amino-salicyclic acid, kanamyin, capreomycin, ethionamide, cycloserine, thiacetazone or a flouroquinolone, or an analogue of such an agent.

50. Use of an antigen or analogue as defined in any one of claims 29 to 47 in the manufacture of a diagnostic means for use in a method according to any one of claims 29 to 47 wherein the method is performed in vivo.

51. A kit for carrying out the method of any one of claims 29 to 47 comprising the antigen or analogue, and optionally also a means to detect whether T cells recognise antigen or analogue.

52. A kit according to claim 51 which also comprises an agent as defined in claim 48 or 49.

53. Method of diagnosing susceptibility to active tuberculosis disease and latent mycobacterial infection in an individual on or about to start immunosuppressive therapy comprising detecting whether or not the T cells of the individual recognise mycobacterial antigen, wherein recognition of mycobacterial antigen by the T cells indicates susceptibility to active tuberculosis disease and latent mycobacterial infection.

54. Method of monitoring susceptibility to active tuberculosis disease and latent mycobacterial infection in an individual on immunosuppressive therapy comprising detecting whether or not the T cells of the individual recognise mycobacterial antigen, wherein recognition of mycobacterial antigen by the T cells indicates susceptibility to

active tuberculosis disease and latent mycobacterial infection.

55. Method according to claim 53 or 54 wherein the immunosuppressive therapy comprises administration of an anti-TNF-∀ antibody, methotrexate, azathioprine, a corticosteroid or mycophenolate mofetil .

56. Method according to claim 54 wherein the antibody is infliximab.

57. Method according to any one of claims 53 to 56 wherein T cell recognition is performed using a method as defined in any one of the preceding claims.

58. Method according to any one of claims 53 to 57 comprising determining in vitro whether the T cells of the individual recognise/react to a mycobacterial antigen or an analogue of the mycobacterial antigen.

59. Method according to any one of claims 53 to 58 comprising contacting a population of T cells from the individual with (a) one or more peptide epitopes of a mycobacterium, or (b) an analogue of said peptide(s) which is recognised by T cells that recognise said peptide(s), and determining the reaction of the T cells to the peptide(s) or analogue(s).

60. Method according to any one of claims 53 to 59 in which the peptide epitope, or the analogue of the peptide epitope, has a length of 8 to 100 amino acids.

61. Method according to any one of claims 53 to 60 wherein whether or not the T cells recognise a pool of a least 4 peptide epitopes, or analogues thereof, is determined.

62. Method according to any one of claims 53 to 61 in which during detection of the reaction of the T cells to the peptide, or the analogue of the peptide, antigen presenting cells are present which are capable of processing the peptide or analogue and presenting it to the T cells.

63. Method according to any one of claims 53 to 62 wherein the mycobacterial antigen is of M. tuberculosis.

64. Method according to any one of claims 53 to 63 wherein the mycobacterial antigen is ESAT-6 or CFP10.

65. Method according to any one of claims 53 to 64 wherein recognition by the T cells of a peptide or peptides chosen from one or more of the following peptide epitopes is determined:

MTEQQWNFAGIEAAA
WNFAGIEAAASAIQG
IEAAASAIQGNVTSI
SAIQGNVTSIHSLLD
NVTSIHSLLDEGKQS
HSLLDEGKQSLTKLA
EGKQSLTKLAAAWGG
LTKLAAAWGGSGSEA
AAWGGSGSEAYQGVQ
SGSEAYQGVQQKWDA
YQGVQQKWDATATEL
QKWDATATELNNALQ
TATELNNALQNLART
NNALQNLARTISEAG
NLARTISEAGQAMAS
ISEAGQAMASTEGNV
QAMASTEGNVTGMFA
MAEMKTDAATLAQEA
TDAATLAQEAGNFER
LAQEAGNFERISGDL
GNFERISGDLKTQID
ISGDLKTQIDQVEST

KTQIDQVESTAGSLQ
QVESTAGSLQGQWRG
AGSLQGQWRGAAGTA
GQWRGAAGTAAQAAV
AAGTAAQAAVVRFQE
AQAAVVRFQEAANKQ
VRFQEAANKQKQELD
AANKQKQELDEISTN
KQELDEISTNIRQAG
EISTNIRQAGVQYSR
IRQAGVQYSRADEEQ
VQYSRADEEQQQALS
ADEEQQQALSSQMGF

or an analogue thereof which is recognised by a T cell which recognises the peptide epitope.

66. Method according to any one of claims 53 to 65 wherein T cell recognition is determined by detecting secretion of a cytokine from the T cells.

67. Method according to claim 66 in which the cytokine is IFN-(.

68. Method according to claim 66 or 67 in which the cytokine is detected by allowing the cytokine to bind to an immobilised antibody specific to the cytokine and then detecting the presence of the antibody/cytokine complex.

69. Method of treating an individual comprising administering to an individual diagnosed as being susceptible to active tuberculosis disease or latent mycobacterial infection by a method according to any of claims 53 to 68, a product which prevents or treats said disease or infection.

70. Method according to claim 69 wherein the product is rifampicin, isoniazid, pyrazinamide, ethambutol, streptomycin, para-amino-salicyclic acid, kanamyin,

capreomycin, ethionamide, cycloserine, thiacetazone or a flouroquinolone, or an analogue of such a product.

71. A kit for carrying out the method of any one of claims 53 to 68 comprising a mycobacterial antigen or an analogue thereof, and optionally also a means to detect whether T cells recognise the mycobacterial antigen or analogue.

72. A kit according to claim 70 wherein the mycobaterial antigen or analogue thereof comprises a peptide epitope which is 8 to 100 amino acids longs.

73. A kit according to claim 71 or 72 which also comprises a product as defined in claim 69 or 70.

74. Method of detecting onset of active mycobacterial disease, or susceptibility to onset of active mycobacterial disease, in an individual who does not have any symptoms of mycobacterial disease comprising determining whether the individual has increased levels of T cells which recognise a mycobacterial antigen, to thereby determine onset of mycobacterial disease or whether the individual is susceptible to onset of active mycobacterial disease.

75. Method according to claim 74 comprising determining whether the level of said T cells in the individual is at least five-fold higher compared to a previous time point.

76. Method according to claim 74 or 75 comprising determining whether the level of said T cells in the individual is at least 100 per million peripheral blood mononuclear cells higher compared to a previous time point.

77. Method according to any one of claims 74 to 76 comprising determining at a first time point the basal level of mycobacterial antigen specific T cells in the individual and determining at a subsequent second time point whether or not the level of said T cells in the individual has increased.

78. Method according to claim 77 wherein the first and second time point are separated by about 3 to 24 months.

79. Method according to any one of claims 74 to 78 wherein the level of T cells is detected by measuring in vitro the recognition/reaction of the T cells to a mycobacterial antigen or an analogue of the mycobacterial antigen.

80. Method according to any one of claims 74 to 79 comprising contacting a population of T cells from the individual with (a) one or more peptide epitopes of a mycobacterium, or (b) an analogue of said peptide(s) which is recognised by T cells that recognise said peptide(s), and determining the reaction of the T cells to the peptide(s) or analogue(s).

81. Method according to any one of claims 74 to 80 in which the peptide epitope, or the analogue of the peptide epitope, has a length of 8 to 100 amino acids.

82. Method according to any one of claims 74 to 81 wherein the reaction of the T cells to a pool of a least 4 peptide epitopes, or analogues thereof, is determined.

83. Method according to any one of claims 74 to 82 in which during detection of the reaction of the T cells to the peptide, or the analogue of the peptide, antigen presenting cells are present which are capable of processing the peptide or analogue and presenting it to the T cells.

84. Method according to any one of claims 74 to 83 wherein the mycobacterial antigen is of M. tuberculosis.

85. Method according to any one of claims 74 to 84 wherein the mycobacterial antigen is ESAT-6 or CFP10.

86. Method according to any one of claims 74 to 85 wherein recognition by the T cells of a peptide or peptides chosen from one or more of the following peptide epitopes is

determined:

MTEQQWNFAGIEAAA
WNFAGIEAAASAIQG
IEAAASAIQGNVTSI
SAIQGNVTSIHSLLD
NVTSIHSLLDEGKQS
HSLLDEGKQSLTKLA
EGKQSLTKLAAAWGG
LTKLAAAWGGSGSEA
AAWGGSGSEAYQGVQ
SGSEAYQGVQQKWDA
YQGVQQKWDATATEL
QKWDATATELNNALQ
TATELNNALQNLART
NNALQNLARTISEAG
NLARTISEAGQAMAS
ISEAGQAMASTEGNV
QAMASTEGNVTGMFA
MAEMKTDAATLAQEA
TDAATLAQEAGNFER
LAQEAGNFERISGDL
GNFERISGDLKTQID
ISGDLKTQIDQVEST
KTQIDQVESTAGSLQ
QVESTAGSLQGQWRG
AGSLQGQWRGAAGTA
GQWRGAAGTAAQAAV
AAGTAAQAAVVRFQE
AQAAVVRFQEAANKQ
VRFQEAANKQKQELD
AANKQKQELDEISTN

KQELDEISTNIRQAG
EISTNIRQAGVQYSR
IRQAGVQYSRADEEQ
VQYSRADEEQQQALS
ADEEQQQALSSQMGF

or an analogue thereof which is recognised by a T cell which recognises the peptide epitope.

87. Method according to any one of claims 74 to 86 wherein T cell recognition is determined by detecting secretion of a cytokine from the T cells.

88. Method according to claim 87 in which the cytokine is IFN-(.

89. Method according to claim 87 or 88 in which the cytokine is detected by allowing the cytokine to bind to an immobilised antibody specific to the cytokine and then detecting the presence of the antibody/cytokine complex.

90. Method of treating an individual comprising administering to an individual diagnosed as being susceptible to active tuberculosis disease or latent mycobacterial infection by a method according to any of claims 74 to 89, a product which prevents or treats said disease or infection.

91. Method according to claim 90 wherein the product is rifampicin, isoniazid, pyrazinamide, ethambutol, streptomycin, para-amino-salicyclic acid, kanamyin, capreomycin, ethionamide, cycloserine, thiacetazone or a flouroquinolone, or an analogue of such a product.

92. A kit for carrying out the method of any one of claims 74 to 91 comprising a mycobacterial antigen or an analogue thereof, and optionally also a means to detect whether T cells recognise the mycobacterial antigen or analogue.

93. A kit according to claim 92 wherein the mycobaterial antigen or analogue thereof

comprises a peptide epitope which is 8 to 100 amino acids longs.

94. A kit according to claim 92 or 93 which also comprises a product as defined in claim 90 or 91.